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Caloplaca allochroa (lichenized *Ascomycetes*), a new saxicolous lichen species from South Korea

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Abstract — The saxicolous lichen species, *Caloplaca allochroa,* is described as new from subtropical inland localities of Gangwon and Jeollanam Province (South Korea). It is characterized by occurring as two colour variants, yellowish and gray, based on presence vs. absence of anthraquinone pigments. Its colour contrast between brownish-red to brownish-black apothecial disc and yellowish-orange apothecial margin is very characteristic. Further diagnostic characters are a hymenium with numerous oil-drops and specific ecology. The species is related to *Caloplaca flavorubescens* and *C. flavovirescens.*

Key words — anthraquinones, ITS nrDNA, phylogeny, taxonomy, *Teloschistaceae*

Introduction

A study of specimens of the genus *Caloplaca* Th. Fr. collected during field trips in the mountains of South Korea by L. Lőkös and Y. Joshi, revealed an interesting and unusual saxicolous species, which is described here as new to science. The new species has been found only at inland localities in three mountains, Mt Jobong, Mt Taebaek, and Mt Gyeokja. It grows in open woodlands on siliceous and non-calcareous rocks together with abundant members of *Aspicilia*, *Cladonia*, *Heterodermia*, *Lecanora*, *Leptogium*, *Myelochroa*, *Pertusaria*, *Phaeophyscia*, *Porpidia,* and *Ramalina*.

Although we obtained only three specimens of this new taxon, we describe it without any hesitation because 1) the species is morphologically well defined,

2) one may wait for too a long time to obtain more material of this rare taxon when conservationists and scientists could benefit from the data, 3) DNA evidence supports our hypothesis, and 4) the material was collected in ample amount and duplicates sent to several herbaria.

Materials & methods

Studied specimens are deposited in CBFS, KH, KoLRI, UCR and the private herbarium of J. Halda (Czech Republic). Description and photographs of external morphology are based on air-dried material observed under a dissecting stereomicroscope (Nikon SMZ645). Sections were made with a razor blade under the stereomicroscope and mounted in lactophenol cotton blue. Anatomical descriptions are based on these preparations under a compound microscope (Nikon Eclipse E200). The mycological terminology follows Kirk et al. (2008). Measurements of ascospores were made at ×400 magnification mounted in water; only free ascospores lying outside the asci were measured. The results are recorded as 95% confidence interval of the measured values. Number of measurements (n) is within square brackets. Chemical constituents were identified by HPLC (Søchting 1997).

DNA extraction and amplification. Direct PCR was used for PCR-amplification of the ITS regions including the 5.8S gene of the nuclear rDNA following Arup (2006). Primers for amplification were ITS1F (Gardes & Bruns 1993) and ITS4 (White et al. 1990). PCR cycling parameters follow Ekman (2001).

Phylogenetic analysis. Five newly obtained ITS sequences were included in the phylogenetic analysis along with six sequences from GenBank (Tab. 1) to illustrate the phylogenetic position of *Caloplaca allochroa* within the *Caloplaca flavorubescens*/*C. flavovirescens* clade. *Caloplaca crenularia* and *C. ferruginea* were selected as outgroup. Sequences were aligned using BioEdit and manually cut to eliminate the unaligned

Table 1. Sequences used in phylogenetic analysis.*

Species	Voucher data	Genbank accession number
Caloplaca allochroa Y. Joshi, Vondrák & Hur	South Korea; CBFS JV7987	**HQ415800**
C. crenularia (With.) J.R. Laundon	Arup et al. 2007	EF643512
C. egeana Cl. Roux & Nav.-Ros.	Gibraltar; CBFS JV6262	**HQ644198**
C. ferruginea (Huds.) Th. Fr.	Arup & Åkelius 2009	FJ866808
C. flavorubescens (Huds.) J.R. Laundon	Wei et al. 2008	EU266111
	Wedin et al. 2002	AY143394
	Kasalicky et al. 2000	AF279887
C. flavovirescens (Wulfen) Dalla Torre & Sarnth.	Arup & Grube 1999	AF353966
C. juniperina Tomin	Tajikistan; CBFS JV7397	**HQ644200**
	Tajikistan; herb. Halda 174	**HQ644199**
C. stantonii W.A. Weber ex Arup	California; UCR 55976	**HQ644197**

* Newly obtained sequences shown in bold.

ends and ambiguously aligned regions of ITS1 and ITS2; 539 positions were retained. Bayesian phylogenetic analysis was carried out using the program MrBayes 3.1.1 (Ronquist & Huelsenbeck 2003). The optimal nucleotide substitution model (GTR+G) was found using the program MrModeltest v2.3 (Nylander 2004) with the Akaike Information Criterion and the hierarchical likelihood ratio test (Posada & Crandall 1998). The MCMC analysis was run for one million generations, performed in two runs, each with four chains starting from a random tree and using the default temperature of 0.2. Every 100th tree was sampled, and the first 215,000 generations were discarded as burn-in, using standard deviation of splits between runs less than 0.01 as a convergence criterion.

New species

Caloplaca allochroa Y. Joshi, Vondrák & Hur, **sp. nov.** Fig. 1

MycoBank MB 561554

Thallus crustaceus, continuus ad rimosus ad disperso-areolatus, laevigatus, tenuis, luteus vel cinereus, UV–. Prothallus praesentia, nigrescens. Apothecia biatorina, sessilia, rotundata ad angulatus, 0.3–0.8 mm. Discus planiusculus ad ±convexus, porphyreus ad fuscus, ±pruinosus. Proper margine flavoaurantiacus, persistens. Hymenium inspersum. Hypothecium indistinctus cellularis. Proper margine tenuis, prosoplectenchymatous. Thalline margine absentia. Paraphyses simplices, ±furcatus ad apicis, septatus. Asci 8-spori, ascosporae polarilocularis, ellipsoideus ad late ellipsoideus, 15–20 µm longae et 7.5–10 µm latae.

Type: South Korea. Gangwon Prov.: Yangyang-gun, Seo-myeon, Hwang-ri, Mt Jobong, 37°56′10″N, 128°33′74″E, alt. 980 m, on siliceous rock, 14 May 2009, Y. Joshi, X.Y. Wang, J.A. Ryu, J.Y. Hur 090296 (**holotype** KoLRI; **isotypes**, CBFS JV7987, KH).

Etymology — The species epithet derives from the Latin word *allochrous,* which means changing colour.

Thallus saxicolous, crustose, continuous to rimose to dispersed areolate, smooth, indeterminate, effuse, 6–8 cm in diam., 70–90 µm thick [n = 20], whitish-gray (Fig. 1A) or yellow (Fig. 1B). Cortex thin to moderately thick, 8–23 µm thick [n = 20], paraplectenchymatous, necral layer absent. Algal layer continuous, 25–50 µm high [n = 20]. Medulla hyaline, of densely interwoven hyphae, without crystals, 12.5–40 µm high [n = 20]. Prothallus blackish-gray.

Apothecia biatorine, numerous, scattered to ±aggregated, 0.3–1.0 mm in diam. [n = 20], sessile to ±constricted at the base, slightly concave when young, later plane to slightly convex; disc rounded to irregular, brownish-red to brownish-black, ± greenish-yellow pruinose (Fig. 1B); margin egg yolk coloured to yellowish-orange (distinctly paler than disc) to orange-brown, variously thick, flush, smooth to ±wavy, entire, persistent. Epihymenium dark golden brown, with numerous yellowish brown granules, 12.5–20 µm high [n = 20]. Hymenium hyaline, with numerous oil droplets not dissolving in K, 87.5–100 µm high [n = 20]. Hypothecium hyaline, of indistinct

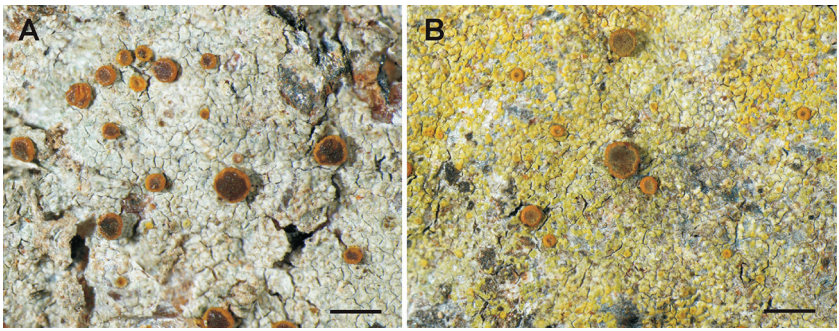

Fig. 1 *Caloplaca allochroa* (isotype). **A.** White-gray thallus with apothecia (Scale = 1 mm); **B.** Yellow thallus with apothecia (Scale = 1 mm).

cells, 75–110 µm [n = 20]. Proper margin thin to thick, 25–75 µm wide [n = 20], prosoplectenchymatous, with outer region yellowish-brown pigmented. Algal layer ±continuous below the hypothecium. Thalline margin absent. Paraphyses hyaline, simple to ±furcated at the apices, septate, 1.5–2 µm thick, tips 2.5–3 µm broad [n = 20]. Ascospores 8 per ascus, hyaline, polarilocular, ellipsoid to broadly ellipsoid, 12.5–21.5 × 6–11.5 µm, isthmus 2.5–7.5 µm [n = 20]. Pycnidia rarely present, ostioles reddish-orange; conidia bacilliform, 2.1–2.5 × 0.7–0.9 µm [n = 20].

Chemistry — Spot test reactions: thallus K– (grayish regions) or K+ purple (yellowish regions), C–, KC–, P–, UV–. Epihymenium K+ purple, C–. Apothecia contain parietin and fragilin (major anthraquinones), emodin and 7-chloroemodin (traces); fragilin was observed as the major pigment in yellow thalli together with traces of parietin, emodin and 7-chloroemodin. Only traces of parietin and fragilin were observed in white thalli. No acetone insoluble pigments were identified in darkened apothecial discs.

Phylogeny — *Caloplaca allochroa* is placed in the clade containing well-known species *C. flavorubescens* (Huds.) J.R. Laundon and *C. flavovirescens* (Wulfen) Dalla Torre & Sarnth. (PP=1.00). The clade is not internally well-resolved with *C. egeana*, *C. flavovirescens* and *C. stantonii* in the basal polytomy (Fig. 2).

Ecology and distribution — *Caloplaca allochroa* is so far known only from three mountains (Mt Jobong, Mt Taebaek, and Mt Gyeokja) in South Korea. The species occurs on non-calcareous rocks in subtropical inland areas at elevations from 350–980 m. It grows both on subvertical and horizontal faces of the rocks, preferring situations with sufficiency of diffuse sunlight, under open canopied stands. Both yellow and gray-white morphotypes are reported from Mt Jobong and Mt Gyeokja, while in Mt Taebaek only the gray-white morphotype has been collected.

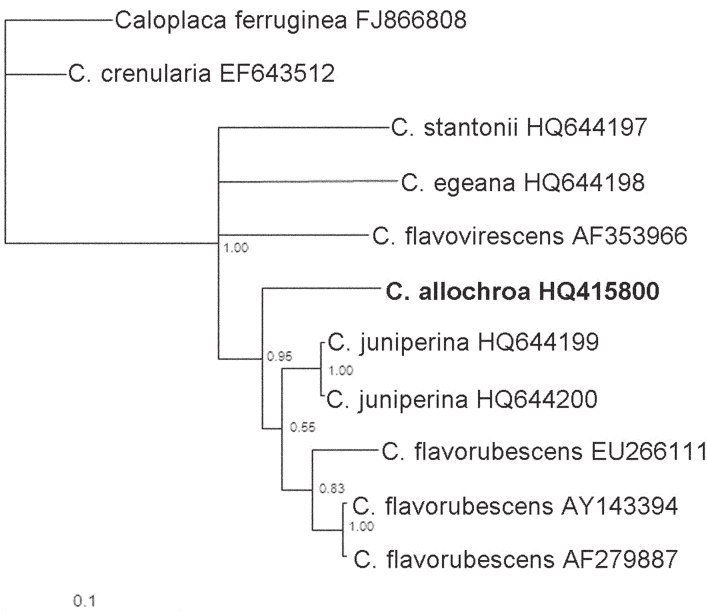

Fig. 2 Bayesian phylogenetic tree based on ITS nrDNA sequences showing the alliance of *Caloplaca allochroa* with *C. flavorubescens* and *C. flavovirescens*. Posterior probabilities are present at nodes.

Its three known communities have rather different lichen compositions. At the type locality *Caloplaca allochroa* was growing in a community with species of *Cladonia, Heterodermia, Leptogium, Myelochroa, Pertusaria, Porpidia* and *Ramalina*. The new species was accompanied only by *Aspicilia* sp. on Mt Taebaek but by *Leptogium, Phaeophyscia, Myelochroa, Pertusaria,* and *Lecanora* on Mt Gyeokja. Even though phorophytes, such as *Acer, Fraxinus, Rhododendron, Pinus,* and *Quercus* occur in all three *C. allochroa* localities, the new taxon has not yet been found growing on bark.

Additional specimens examined: **SOUTH KOREA**. **Gangwon Prov.**: Taebaek-si, Mungoksodo-dong, Mt Taebaek, 37°06′08.3″N, 128°57′05.4″E, alt. 970 m, on rock, 14 October 2005, L. Lőkös 050734 (KoLRI). **Jeollanam Prov.**: Wando Co., Bogil-myeon, Bogil Island, Buyong-ri, Mt Gyeokja, en route from Keungiljae to Suribong, 34°08′50″N, 126°32′90″E, alt. 368 m, on rock, 05 February 2010, Y. Joshi, H. S. Jeon & M . H. Jeong 100152 (KoLRI).

Remarks — This is a distinctive species characterized by the colour variation shown by its thallus (yellow to gray), dark brown-red biatorine apothecia with yellow-orange margin, and inspersed hymenium. The colour of the areoles is

yellow and the discs are orange brown without traces of black colour in less sun-exposed sites but on the strongly exposed rock faces, the thallus is distinctly gray and the apothecia are brownish-black.

The yellow morphotype of the new species is most likely to be confused with the saxicolous *Caloplaca flavovirescens* or the corticolous *C. alnetorum* Giralt et al., *C. flavorubescens,* and *C. gordejevii* (Tomin) Oxner ex Khodos., but none of the latter possesses such a strong colour contrast between apothecial disc and margin. Gray-white thallus morphotypes are similar to saxicolous *Caloplaca egeana* Cl. Roux & Nav.-Ros. and *C. subochracea* (Wedd.) Werner, and corticolous *C. aegatica* Giralt et al., which also differ in apothecial coloration. *Caloplaca lypera* Poelt & Hinter., a similar species from temperate or sub-alpine Himalayan regions, cannot be confused with this taxon, as it differs in having yellowish medulla and ochraceous-gray thallus (Poelt & Hinteregger 1993; Joshi & Upreti 2011). The combination of parietin and fragilin as main anthraquinones in the new species is rare within *Caloplaca*; it is known (as in *C. flavovirescens* and *C. flavorubescens;* Santesson 1970) and described as chemosyndrome B1 by Søchting (2001).

Existence of two colour thallus variants based on presence vs. absence of anthraquinones is probably not exceptional in *Teloschistaceae*; it has been recently described in *Caloplaca phlogina* (Ach.) Flagey (Vondrák et al. 2010) and also occurs in the North American *C. stanfordensis* H. Magn. (Vondrák, unpublished data).

Acknowledgments

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